

***Pseudocercospora seropedicensis* & *P. solani-cernui* spp. nov. on *Solanum* from Brazil**

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ABSTRACT—Using stereo- and light microscopy, two leaf-spotting cercosporoid fungi, collected from areas of the Brazilian Atlantic Forest in the states of Minas Gerais and Rio de Janeiro, were shown to represent two new species (*Pseudocercospora seropedicensis* on *Solanum asperum* and *P. solani-cernui* on *S. cernuum*), which are herein described and illustrated.

KEY WORDS—cercosporoid fungi, hyphomycetes, morphology, mycodiversity, *Solanaceae*

Introduction

Cercosporoid fungi, which are found worldwide, are especially abundant and diverse in tropical and subtropical areas of Africa, Asia, Australia, and Central and South America (Beilharz 2002; Braun & Freire 2003, 2004, 2006; Castañeda & Braun 1989; Hernández-Gutiérrez & Dianese 2008, 2009; Inácio & al. 1996; Silva & al. 2016). These fungi have been studied by several authors in Brazil (Batista & al., cf. Silva & Minter 1995; Braun & Freire 2004; Chupp 1954; Crous & Braun 2003; Crous & Wingfield 1997; Crous & al. 1999; Fernandes & al. 2013; Hernández-Gutiérrez & Dianese 2008, 2009; Hernández-Gutiérrez & al. 2014, 2015; Inácio & Dianese 1998, 1999, 2006; Inácio & al. 1996; Pereira & Barreto 2006; Silva & al. 2016; Viégas 1945). They have been reported as plant pathogens on several hosts including different economically important crops,

and some cercosporoid fungi are also used for biological control of weeds (Farr & Rossman 2019, Groenewald & al. 2013, Hanada & Gasparotto 2002, Mendes & Urben 2019, Mendes & al. 1998, Pereira & Barreto 2006, Pons 1987, Spegazzini 1910).

Pseudocercospora Speg. (introduced by Spegazzini 1910 for the type species *P. vitis* (Lév.) Speg.) contains endophytic, pathogenic, or saprotrophic species, some of which are used for weed control (Breeÿen & al. 2006). The genus now comprises synnematal fungi previously assigned to *Phaeoisariopsis* Ferraris (Ferraris 1909) and fungi with slightly pigmented conidiogenous structures and with paracercospora-like loci, although most *Pseudocercospora* species are characterized by unthickened, non-pigmented conidiogenous loci and conidial hila (Braun 1995, Deighton 1976). Sexual morphs are unknown for most species, but when present they are mycosphaerella-like, previously allocated in *Mycosphaerella*, now a heterotypic synonym of *Ramularia* Unger (Braun & al. 2013, Crous & al. 2013).

Solanaceae comprises about 3000 species in 150 genera, distributed primarily in tropical and subtropical South America. In Brazil there are 350 species in 32 genera (Souza & Lorenzi 2008). *Solanum*, the most abundant genus, encompasses about 1500 species and occurs in tropical and subtropical areas worldwide (Sarmiento-Silva & al. 2003). Several economically important species also belong in *Solanaceae*: pepper (*Capsicum*), potato (*Solanum tuberosum*), tomato (*S. lycopersicum*), and tobacco (*Nicotiana tabacum*) (Martins & Costa 1999). Cercosporoid fungi occur on *Solanum* species (Silva & al. 2016), and recently Braun (2017) published an identification key for *Pseudocercospora* species on *Solanum* hosts with descriptions and illustrations for some species.

Here we describe and illustrate two new *Pseudocercospora* species found on leaves of *Solanum asperum* and *S. cernuum*.

Materials & methods

Plant material showing necrotic leaf spots was collected during July 2013–October 2017 in the States of Rio de Janeiro (UFRRJ campus in Paracambi Natural Park and the National Forest in Seropédica) and Minas Gerais (town of Liberdade).

Fresh and dried symptomatic leaves were examined with a dissecting microscope to select fungal structures for mounting on slides with Cotton Blue / lactoglycerol or phloxine / KOH glycerol. Material was also hand-sectioned and mounted on slides. Tissues were microscopically observed and photographed using an Olympus BX41 light microscope coupled to a Canon Power Shot ELPH 100 HS camera. Structures were measured using a calibrated ocular micrometer. The exsiccates were registered and deposited in the Plant Pathological Herbarium “Verlande Duarte Silveira” at the Rural University of Rio de Janeiro (UFRJ).

TABLE 1. Selected *Pseudocercospora* species on *Solanum*

SPECIES	HOSTS	DISTRIBUTION	REFERENCES
<i>P. atromarginalis</i>	<i>Solanum</i> spp., <i>S. americanum</i> , <i>S. lycopersicum</i>	Cuba, Taiwan, Korea, USA	Atkinson (1892), Castañeda & Braun (1989), Choi et al. (2015), Deighton (1976), Guo & Hsieh (1995)
<i>P. marcelliana</i>	<i>S. nudum</i> , <i>S. torvum</i> var. <i>hartwegianum</i> , <i>S. micranthum</i>	Spain, Venezuela	Chupp (1954), Crous & Braun (2003), Braun (2017)
<i>P. seropedicensis</i>	<i>S. asperum</i>	Brazil	This work
<i>P. solani-cernui</i>	<i>S. cernuum</i>	Brazil	This work
<i>P. solani-pseudocapsicola</i>	<i>S. pseudocapsicum</i>	Brazil	Silva et al. (2016)
<i>P. solani-torvicula</i>	<i>S. torvum</i>	Taiwan	Guo & Hsieh (1995)
<i>P. trichophila</i> var. <i>trichophila</i>	<i>Solanum</i> spp.	Asia, Central and South America, North America, Oceania	Stevens (1917), Chupp (1954), Braun (2017)
<i>P. trichophila</i> var. <i>punctata</i>	<i>S. hirtum</i>	Venezuela	Braun & Urtiaga (2012)
<i>P. venezuelae</i>	<i>Solanum</i> sp., <i>S. argenteum</i> , <i>S. "aculeatum"</i> , <i>S. aethiopicum</i> [= <i>S. gilo</i>]	Brazil, Venezuela, South America	Chupp (1954), Crous & Braun (2003), Braun & Freire (2003; 2006), Braun (2017)

Taxonomy

Pseudocercospora seropedicensis Andrade, Medeiros & Inácio, sp. nov. Pl. 1

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Differs from *Pseudocercospora solani-asperi* [$\equiv$ *P. trichophila* var. *trichophila*] by its much longer conidia and conidiophores and its larger, amphigenous, stromata.

TYPE: Brazil, Rio de Janeiro, Seropédica, road to cattle farm, near Instituto de Florestas, 22°45'31"S 43°41'57"W, on leaves of *Solanum asperum* Rich. (*Solanaceae*), 07 May 2016, C. A. Inácio 122, (Holotype, UFRJ 12.390).

ETYMOLOGY: honoring the municipality, Seropédica, where the species was first discovered.

LESIONS ≤ 50 mm diam., epiphyllous, sparse, sometimes gregarious, confluent, circular to irregular, reddish brown to light brown in the center, later becoming necrotic, grayish, with dark brown border. COLONIES amphigenous, mainly hypophyllous, caespitose, sparse. SUPERFICIAL MYCELIUM colorless to pale brown, septate, branched, amphigenous, mostly hypophyllous, sometimes emerging through the stomata, bearing conidiophores; hyphae 2–4(–7) μ m

diam.; CONIDIOPHORES $5\text{--}42(-45) \times 3\text{--}4 \mu\text{m}$, 1-septate, sometimes curved, slightly geniculate due to sympodial proliferation, not branched, borne terminally or (usually) laterally on superficial hyphae, olive to olive brown, rounded at apex. INTERNAL MYCELIUM colorless to olivaceous brown or pale brown, septate, branched, penetrating deep into the mesophyll and spreading through leaf tissue; hyphae $2\text{--}5(-6) \mu\text{m}$ diam. STROMATA $25\text{--}80(-85) \times 20\text{--}140(-160) \mu\text{m}$, brown, amphigenous, subepidermal, erumpent, textura angularis; cells $4\text{--}7 \mu\text{m}$ wide, giving rise to a fascicle of conidiophores; CONIDIOPHORES $20\text{--}70 \times 1\text{--}4 \mu\text{m}$, 1–5-septate, numerous, macronematous, mononematous, olive to olive brownish, smooth, densely caespitose, arising from stromata, straight or slightly sinuous. CONIDIOGENOUS CELLS integrated, polyblastic, usually monoblastic and terminal, sympodial, geniculate, with inconspicuous conidiogenous loci. CONIDIA solitary, $20\text{--}145(-226) \times 2\text{--}6 \mu\text{m}$, 1–16-septate, straight, sometimes sinuous to curved, subcylindrical to obclavate, smooth, hyaline, subhyaline to olivaceous brown, attenuated at apex, obconically truncate at base, hila neither thickened nor darkened, $1\text{--}2 \mu\text{m}$ wide.

COMMENTS—Several *Pseudocercospora* species on solanaceous hosts are morphologically close to *P. seropedicensis*. *Pseudocercospora atromarginalis* (G.F. Atk.) Deighton (Deighton 1976), *P. marcelliana* (Chupp) U. Braun & Crous (Crous & Braun 2003; Braun 2017), *P. solani-torvicola* Goh & W.H. Hsieh, *P. trichophila* var. *trichophila* (F. Stevens) U. Braun [as *P. solani-asperi* (R.E.D. Baker & W.T. Dale) Deighton] (Stevens 1917; Baker & Dale 1951; Chupp 1954; Deighton 1976; Crous & Braun 2003; Braun 2017), *P. trichophila* var. *punctata* U. Braun & Urriaga, *P. venezuelae* (Chupp) Deighton, and *P. solani-pseudocapsicola* Meir. Silva & al. are compared in TABLES 1–2. *Pseudocercospora atromarginalis* lacks superficial mycelia with secondary conidiophores; collections from Taiwan with conidiophores ($10\text{--}65 \times 3\text{--}5 \mu\text{m}/0\text{--}4\text{-septa}$) and close to our specimen differed in lacking stromata (Guo & Hsieh 1995, Choi & al. 2015). *Pseudocercospora marcelliana* differs by shorter conidia ($15\text{--}70 \mu\text{m}$) and conidiophores ($5\text{--}25 \mu\text{m}$), an absence of superficial mycelium, and occurrence on other hosts or in different locations (Braun 2017; Crous & Braun 2003; Chupp 1954, as *Cercospora*). *Pseudocercospora venezuelae* differs by shorter conidiophores ($10\text{--}70 \mu\text{m}$), conidia ($15\text{--}30\text{--}65 \mu\text{m}$), and lacks superficial mycelium; and *P. solani-torvicola* differs by smaller ($\leq 30 \mu\text{m}$) stromata, shorter and wider primary conidiophores ($35\text{--}60 \times 4.0\text{--}5.5 \mu\text{m}$), shorter secondary conidiophores ($10\text{--}25 \mu\text{m}$), and shorter conidia ($20\text{--}115 \mu\text{m}$) with fewer septa (1–11).

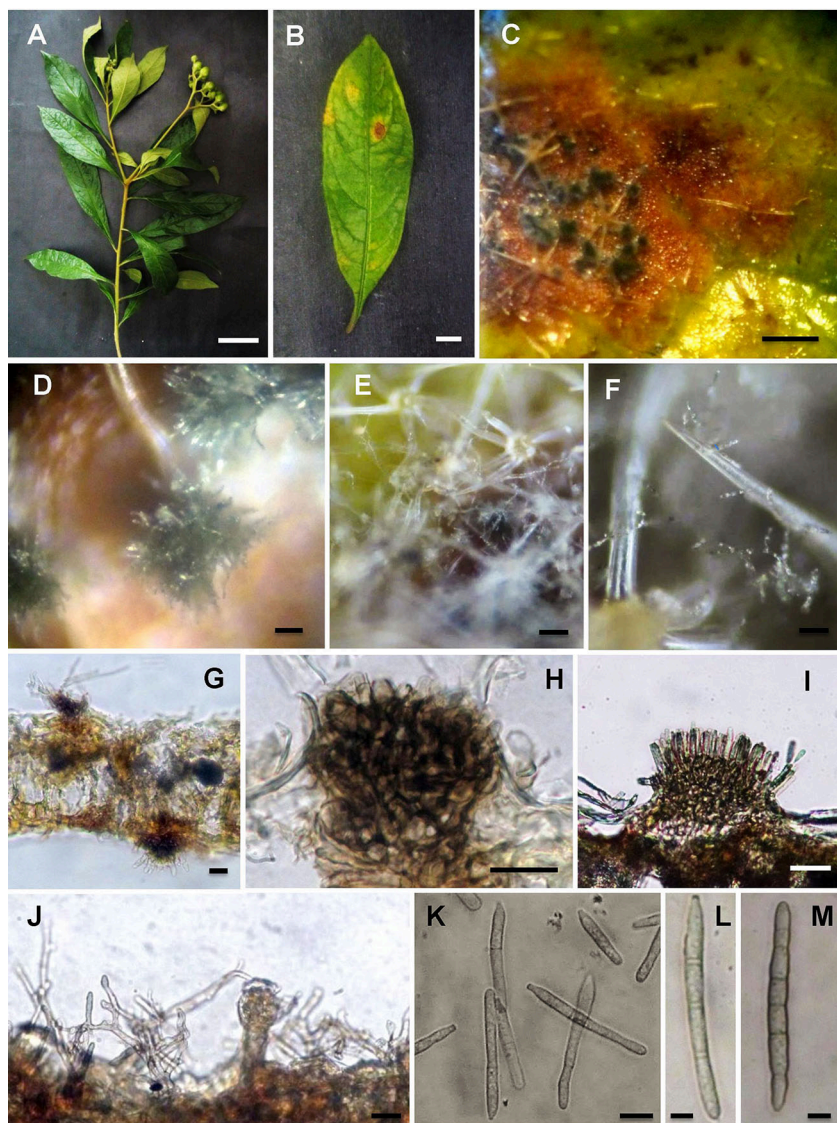


PLATE 1. *Pseudocercospora seropedicensis* (holotype, UFRJ 12.390) on leaves of *Solanum asperum*. A. Branch of *S. asperum*. B. Leaf with lesions. C. Detail of fructifications on lesions. D–F. Conidiophores and conidia as seen under dissecting microscope. G–I. Stroma (vertical section). J. Secondary mycelia on leaf undersurface. K–M. Conidia. Scale bars: A = 5 cm; B = 1 cm; C = 1 µm; D–I = 20 µm; J, K = 10 µm; L, M = 5 µm.

Pseudocercospora trichophila var. *punctata* has smaller dark brown to blackish stromata (10–60 µm diam) (Braun 2017). Finally, although *P. solani-asperi* [≡ *P. trichophila* var. *trichophila*] (Braun 2017) has been reported in Brazil on the same host species (Chupp 1954 as *Cercospora solani-asperi*, Crous & Braun 2003, Deighton 1976 as *Pseudocercospora solani-asperi*, Fernandes & al. 2013), it produces shorter 20–110(165) conidia with fewer 1–10(15) septa; *P. solani-asperi* also usually shows hypophyllous caespituli and lacks a stroma, clearly distinguishing it from the new species found now in the Atlantic Forest. *Pseudocercospora solani-pseudocapsicola*, a new species recently reported on leaves of *Solanum pseudocapsicum* from Brazil (Silva & al. 2016), clearly differs in lacking stromata, shorter (42–128 µm) conidia with fewer (2–6) septa, and hypophyllous conidiophores.

Pseudocercospora seropedicensis is the first report of a *Pseudocercospora* species on leaves of *Solanum asperum* in the State of Rio de Janeiro and the second in Brazil.

ADDITIONAL SPECIMENS EXAMINED—BRAZIL, RIO DE JANEIRO: Paracambi, Parque Natural Municipal do Curió, 22°35'44"S 43°42'18"W, 9.XII.2013, C. A. Inácio 61, (UFRJ 12015); 19.III.2014, C. A. Inácio 74 (UFRJ 12050). Seropédica, Campus UFRRJ, Jardim Botânico near the road to Agricultural Institute, 22°45'53"S 43°41'35"W, 11.VIII.2017, J. M. Oliveira 32 (UFRJ 12.495); 11.VIII.2017, J. D. Almeida 03 (UFRJ 12.496); near Bus Station in front of Female Students Dormitory, 22°45'59"S 43°41'24"W, 22.IX.2017, J. Rembinski 35, J. (UFRJ 12.594); National Forest (Fiona), 22°43'46"S 43°42'36"W, 29.VI.2013, C. A. Inácio 79 (UFRJ 12064).

***Pseudocercospora solani-cernui* Rembinski, Oliveira & Inácio, sp. nov. Pl. 2**

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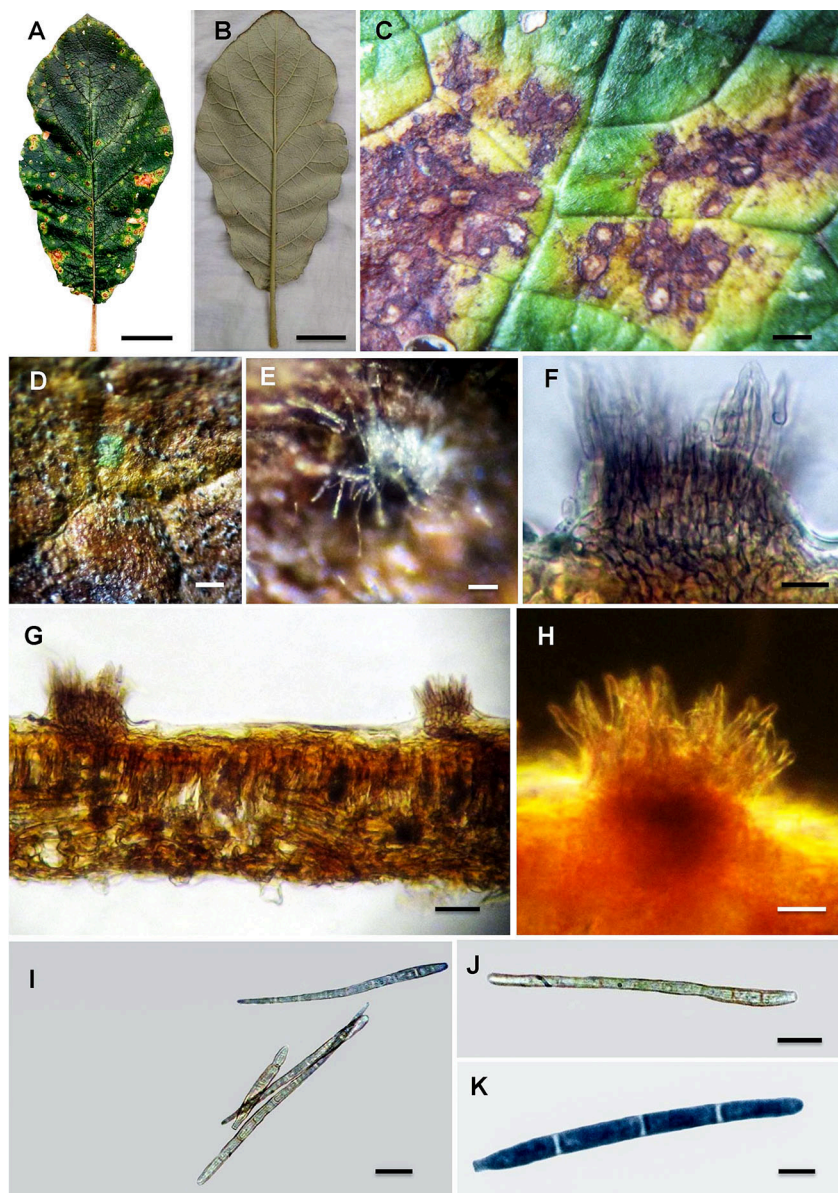
Differs from *P. seropedicensis* by its narrower conidiophores, its larger stromata, and its lack of secondary mycelium.

TYPE: Brazil, Minas Gerais, Liberdade, Sítio Carcará, Zona Rural, Bairro Taquaruçú—Rodovia Joanito Balieiro Km 9, 22°04'26"S 44°23'48"W, on leaves of *Solanum cernuum* Vell. (*Solanaceae*); 14.Oct. 2016, J.C. Oliveira 29 (Holotype, UFRJ 12.422).

ETYMOLOGY: referring to the host species, *Solanum cernuum*.

LESIONS ≤25 mm diam., amphigenous, mainly epiphyllous, sparse, sometimes confluent, circular, later irregular, initially visible as yellowish spots becoming reddish brown to light brown or grayish brown in the center with yellowish margin at the adaxial face and shown as pale areas

PLATE 2. *Pseudocercospora solani-cernui* (holotype, UFRJ 12.286) on leaves of *Solanum cernuum*. A, B. Affected leaf (A: upper surface; B: under surface). C. Leaf spots on upper surface. D. Fructifications on upper surface. E. Conidia and conidiophores seen under stereomicroscope.



F. Stroma with conidiophores showing *textura angularis* (vertical section). G. Stromata (vertical section). H. Stroma and conidiophores. I–K. Conidia. Scale bars: A, B = 5 cm; C = 10 mm; D = 5 mm; E, F = 20 μ m; G = 50 μ m; H = 5 μ m; I, J = 10 μ m; K = 5 μ m.

at the abaxial face of the leaf, becoming slightly brownish at the center, corresponding to the upper leaf spots. COLONIES epiphyllous, caespitose, sparse. MYCELIUM: internal, colorless to pale brown, septate, branched, penetrating the mesophyll; hyphae 2–4 µm diam. STROMATA 10–70 × 30–90(–150) µm, brown, epiphyllous, subepidermal, erumpent, textura angularis, with brownish angular cells; cells 4–7 µm wide, forming a fascicle of conidiophores. CONIDIOPHORES 20–46 × 3–6 µm, 1–3-septate, numerous, macronematous, mononematous, brown, smooth, densely caespitose, arising from stromata, straight or slightly sinuous. CONIDIOGENOUS CELLS integrated, polyblastic, mostly monoblastic and usually terminal, sympodial, geniculate, with inconspicuous conidiogenous loci. CONIDIA solitary, 30–145 × 2–5 µm, 2–12-septate, straight, sometimes sinuous to curved, subcylindrical to obclavate, smooth, subhyaline to brown olivaceous, attenuated at the apex, obconically truncate at the base, hila neither thickened nor darkened, 1–2 µm diam.

TABLE 2. Morphological comparison of selected *Pseudocercospora* species on *Solanum*

SPECIES	STROMATA (µm)	CONIDIOPHORE/ SEPTA (µm)	CONIDIA (µm) / SEPTA	REFERENCES
<i>P. atomarginalis</i>	Absent or only a few brown cells	15–65(100) × 3–5 / 0–4	20–95 × 3.5–5 / 3–11	Guo & Hsieh (1995)
<i>P. marcelliana</i>	15–75	5–30 × 1.5–4 / 1–2	15–110 × 2–3.5 / (1)3–11	Braun (2017)
<i>P. seropedicensis</i>	25–80 (–85) × 20–140 (–160)	1'–20–70 × 1–4 / 1–5; 2'–5–42(45) × 3–5 / 0–1	20–140(226) × 2–6 / 1–16	This work
<i>P. solani-cernui</i>	10–60 × 30–90 (–150)	(15)30–46 × 2–4 / ≤3	30–145 × 2–5 / 2–12	This work
<i>P. solani-pseudocapsicola</i>	Lacking	10–35 × 3–5 / 0–3	42–128 × 2–3.5 / 2–6	Silva et al. (2016)
<i>P. solani-torviculaz</i>	≤30	1'–35–60 × 4.0–5.5 / 1–3 2'–10–25 × 4–5 / 0–1	20–115 × 4.5–6 / 1–11	Guo & Hsieh (1995)
<i>P. trichophila</i> var. <i>punctata</i>	10–60	10–70 × 3–7 / 0–4	(10)20–90(110) × (2.5)3–5 (6) / (1)2–10(12)	Braun & Urtiaga (2012)
<i>P. trichophila</i> var. <i>trichophila</i>	Lacking to few swollen hyphal cells	5–80 × 2.5–5 / 0–4	20–110(165) × 3–5(7) / 1–10(15)	Braun (2017)
<i>P. venezuelae</i>	15–40	10–70 × 2–4 / 0–1(2)	(15–)30–65 × 3–5 / (0–)3–6	Braun (2017)

COMMENTS—*Pseudocercospora solani-cernui* is closely related to *P. trichophila* var. *trichophila*, *P. solani-pseudocapsicola*, and *P. seropedicensis* (TABLES 1–2). Although showing similar conidial dimensions ($20\text{--}110(165) \times 3\text{--}5(7) \mu\text{m}$) and occurring on various *Solanum* species, *P. trichophila* var. *trichophila* is distinguished by much smaller ($\leq 5 \text{ mm}$) leaf spots, absence of a stroma, and longer ($45\text{--}80 \mu\text{m}$) conidiophores (Deighton 1976). *Pseudocercospora solani-pseudocapsicola* (reported on *S. pseudocapsicum* in Brazil) differs in lacking stromata and narrower conidia ($42\text{--}128 \times 2\text{--}3.5 \mu\text{m}$) with fewer (2–6) septa (Silva & al. 2016); *P. seropedicensis* exhibits similar sized stromata ($(20\text{--})140$ (~ 160) μm) but is clearly distinguished from the new species on *S. cernuum* by its much longer ($20\text{--}60(70) \mu\text{m}$) conidiophores, conidia with more (1–16) septa, and superficial mycelium bearing secondary conidiophores.

The above differences morphologically support the *Pseudocercospora* specimen on *S. cernuum* as a new species, here designated as *P. solani-cernui*.

ADDITIONAL SPECIMENS EXAMINED—BRAZIL. RIO DE JANEIRO: Paracambi, Parque Natural Municipal do Curió, $22^{\circ}35'44''\text{S}$ $43^{\circ}42'18''\text{W}$, 05.II.2016, J.M. Oliveira 14 (UFRJ 12.271); 10.VI.2016, J.M. Oliveira 17 (UFRJ 12.290). Seropédica, Campus UFRRJ, reserve in front of the Instituto de Florestas UFRRJ, $22^{\circ}45'30''\text{S}$ $43^{\circ}41'54''\text{W}$, 26.X.2017, C.A. Inácio 160 (UFRJ 12.553). MINAS GERAIS, Liberdade: Sítio Carcará, Zona Rural, Bairro Taquaruçú–Rodovia Joanito Balieiro Km 9, $22^{\circ}04'26''\text{S}$ $44^{\circ}23'48''\text{W}$, 28.X.Oct. 2015, J.C. Oliveira 5 (UFRJ 12172).

Key to *Pseudocercospora* species on *Solanum* from Brazil

- 1 Mycelium internal and external 2
- 1' Mycelium consistently internal 4
- 2 Leaf spots lacking or indistinct, stroma absent *P. trichophila* var. *trichophila*
- 2' Leaf spots amphigenous, stroma well developed *P. seropedicensis*
- 3 Stromata lacking or small as small substomatal clumps of swollen hyphae 4
- 3' Stromata well-developed; often forming sporodochial conidiomata 5
- 4 Conidia $42\text{--}128 \mu\text{m}$ long; on *S. pseudocapsicum* *P. solani-pseudocapsicola*
- 4' Conidia shorter ($\leq 110(120) \mu\text{m}$); on *Solanum* spp. *P. atromarginalis*
- 5 On *Solanum cernuum*; conidia $30\text{--}145 \mu\text{m}$ long, ≤ 12 -septate *P. solani-cernui*
- 5' On *Solanum aculeatum*; conidia $(15\text{--})30\text{--}65 \mu\text{m}$ long, ≤ 6 -septate *P. venezuelae*

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